

# *Chrysochromulina* associated with fish mortalities in a Scottish freshwater loch

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## ABSTRACT

The haptophyte alga *Chrysochromulina* appears to be a common component of freshwater phytoplankton communities in certain Scottish lochs during spring and summer. Toxic events associated with marine *Chrysochromulina* species have been widely reported and confirmed, although direct toxic events associated with fish mortalities in freshwaters are seldom confirmed. Fish mortalities in a freshwater loch in Scotland (Strathclyde Loch) were reported during May 2008 and were associated with abundant *Chrysochromulina*. Although no toxicity tests were carried out, past history of *Chrysochromulina* events in freshwaters should not discount the possibility that this may represent the first report of *Chrysochromulina* toxicity in a Scottish freshwater loch.

## INTRODUCTION

The haptophyta consist of 80 genera and 300 species, with only about a dozen genera known from freshwaters, and only five recorded from the British Isles (Preisig, 2002). *Chrysochromulina* are mostly found in brackish and marine habitats, and although more than 50 species are detailed in total, only 4 freshwater species are known (*C. breviturrita* Nicholls, *C. inornamenta* Wujek & Gardiner, *C. laurentiana* Kling, and *C. parva* Lackey), with *C. parva* being the only one reported from the British Isles (Preisig, 2002).

Although freshwater *Chrysochromulina* appear to be widely distributed within the British Isles (Parke *et al.*, 1962; Lund, 1961), the small and fragile nature of the cell may lead to it not being so easily identified, especially in samples preserved with Lugol's iodine. In Scotland, *Chrysochromulina* has been detected in a number of Scottish freshwater lochs during spring and during late summer (SEPA unpublished data).

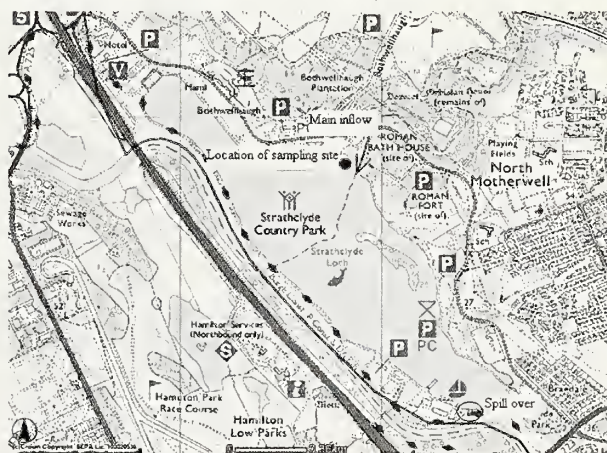
Toxic events associated with blooms of *Chrysochromulina* in marine waters have been extensively reported leading to extensive fish mortalities. There are relatively few toxic reports associated with *Chrysochromulina* in fresh waters. In north American lakes, *C. breviturrita* was suspected as being toxic to tadpoles and producing obnoxious odours (Nichols *et al.*, 1982), whereas dead fish (roach, perch and pike) associated with *C. parva* were observed in a small Danish lake (Hansen *et al.*, 1994). Very high densities of *Chrysochromulina* were also reported

from a small lake in Lincolnshire, England during 1997, which was associated with an extensive fish mortality (SJ Brierley pers. comm). No associated fish mortalities, however, have so far been reported from any Scottish freshwater loch.

## METHODS

On May 6, 2008, following a period of warm, sunny weather, fish mortalities were reported from Strathclyde Loch (North Lanarkshire, Scotland, NS 7269 5734). The man-made loch is approximately 0.8km<sup>2</sup>, situated 20km south-east from Glasgow, Scotland, and is used for a range of recreational activities. The main inflow to the loch comes from the South Calder Water in the north-eastern part, with the main outflow/spillover to the River Clyde in the southern end of the loch.

Water samples were taken at a site in the north-eastern part of the loch (Fig. 1), as far away from the shore as practical, for chemical and biological (phytoplankton) analyses and were analysed following standard Scottish Environment Protection Agency procedures. Additional water samples were taken from the same site a week later, and continued at roughly 2 day intervals, terminating on 27 May. One additional sample was also taken on June 3. On these subsequent surveys, live phytoplankton were immediately examined (inverted microscope method, Leica DMIRB, up to x640 magnification), whereas additional samples were also preserved with Lugol's iodine solution for microscopic enumeration and biovolume assessment (Carvalho *et al.*, 2007).



**Fig 1.** Strathclyde Loch showing sampling site.

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## RESULTS

In the period from 6<sup>th</sup> May up to mid May 2008, approximately 2000 small fish (less than 10cm long) and between 40-50 larger fish (23cm long), mainly roach (*Rutilus rutilus*) and bream (*Abramis brama*), were found dead washed up on the shore. The loch contains a mixture of other fish, such as perch (*Perca fluviatilis*), pike (*Esox lucius*), tench (*Tinca tinca*) and

stickleback (*Gasterosteus aculeatus*), which appeared to have been unaffected, although two dead pike were also found washed up on the shore. On initial and further inspection, no evidence of pollution was indicated, and an ecological survey indicated no dead or dying invertebrates. Water chemistry results indicated relatively high dissolved oxygen concentrations (highest 175%saturation), temperature (19°C) and pH 8.6, which declined towards the end of the sampling period (Table 1).

Determinant	6 May	16 May	28 May	3 June
Dissolved oxygen %saturation	136	175	93.5	-
Temperature °C	19	16	13	-
pH	8.6	-	-	8.44
Conductivity µS/cm	617	-	-	601
Total oxidised nitrogen mg/l	0.412	-	-	0.747
Ammoniacal nitrogen mg/l	<0.04	-	-	0.058
Orthophosphate mg/l	0.128	-	-	0.018

**Table 1.** Selected water chemistry results, Strathclyde Loch, site near the South Calder inflow.

	Relative abundance
Cyanobacteria	
<i>Aphanothece cf. clathrata</i> W. et GS West	+
Centric diatoms	
<i>Stephanodiscus hantzschia</i> Grunow in Cleve et Grunow 1880	+++
Pennate diatoms	
<i>Diatoma tenue</i> var. <i>elongatum</i> Lyngb.1819	+
<i>Nitzschia acicularis</i> (Kütz) W.Sm 1853	+
Cryptophyta	
<i>Cryptomonas</i> spp.	++
<i>Rhodomonas lacustris</i> var. <i>nannoplantica</i> (Skuja) Jav.	++
Chlorophyta/Chlorococcales	
<i>Lagerheima genevensis</i> (Chodat) Chodat	+
<i>Monoraphidium contortum</i> (Thur) Komárková-Legnerová	+
<i>Monoraphidium griffithii</i> (Berk) Komárková-Legnerová	+
<i>Scenedesmus communis</i> Hegewald	+
Chlorophyta/Klebsormidiales	
<i>Koliella longiseta</i> (Vischer) Hindák 1963	+
cf. heterotrophic flagellates	++
Haptophyta	
<i>Chrysochromulina parva</i> Lackey 1939	+++
+ present, ++ common, +++ abundant	

**Table 2.** Dominant phytoplankton recorded 7 May 2008, Strathclyde Loch.



Examination of live phytoplankton on 7 May indicated an abundance of the diatom *Stephanodiscus hantzschia* along with unidentified flagellate nanophytoplankton, which on subsequent examinations were identified and confirmed as *C.parva*. The dominant phytoplankton recorded are shown in Table 2.

*C. parva* abundance declined over May from maxima of 11,500 cells/ml to its lowest by June (15 cells/ml). The biovolume however remained relatively

unchanged through most of early May (between  $0.36 \times 10^6 - 0.48 \times 10^6 \mu\text{m}^3/\text{ml}$ ), and only declined by end May (Figs. 2-3).

Diatom abundance and biovolume fluctuations were similar, with abundance declining from its maximum of 34,800 cells/ml to its lowest by June (153 cells/ml), whereas the biovolume declined from  $5.9 \times 10^6$  to  $0.02 \times 10^6 \mu\text{m}^3/\text{ml}$  over the same period.

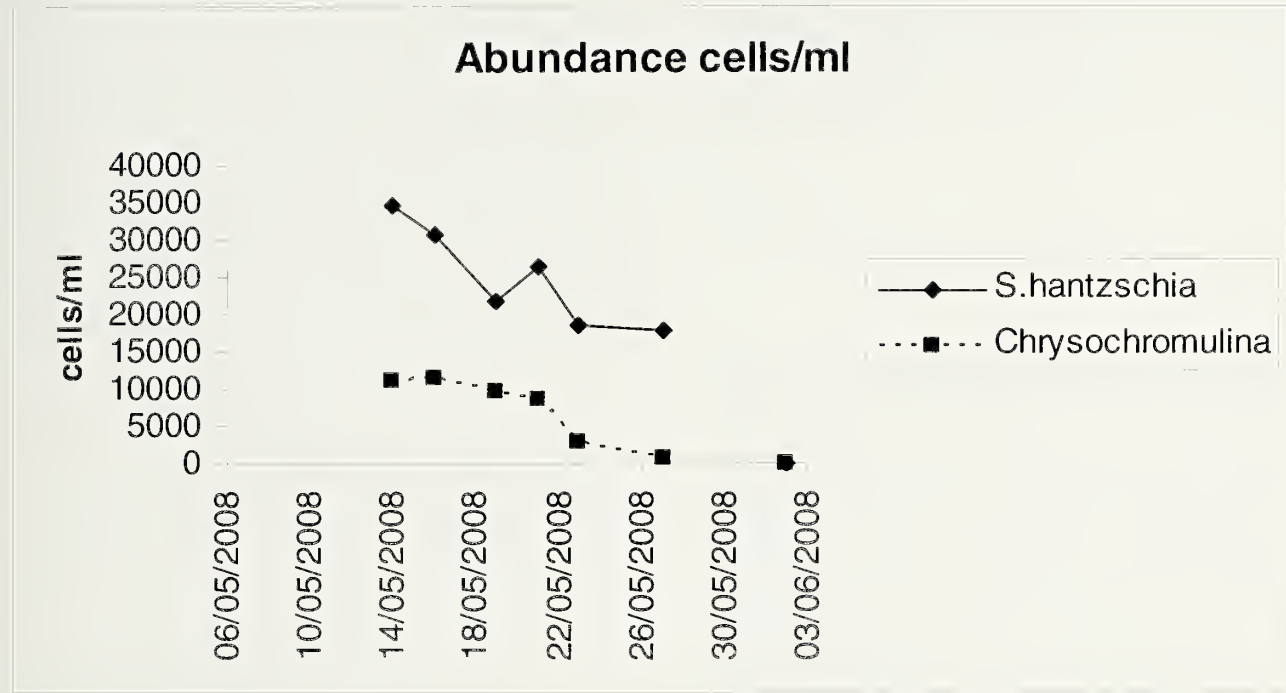


Fig. 2. Abundance of the two dominant algae in Strathclyde Loch.

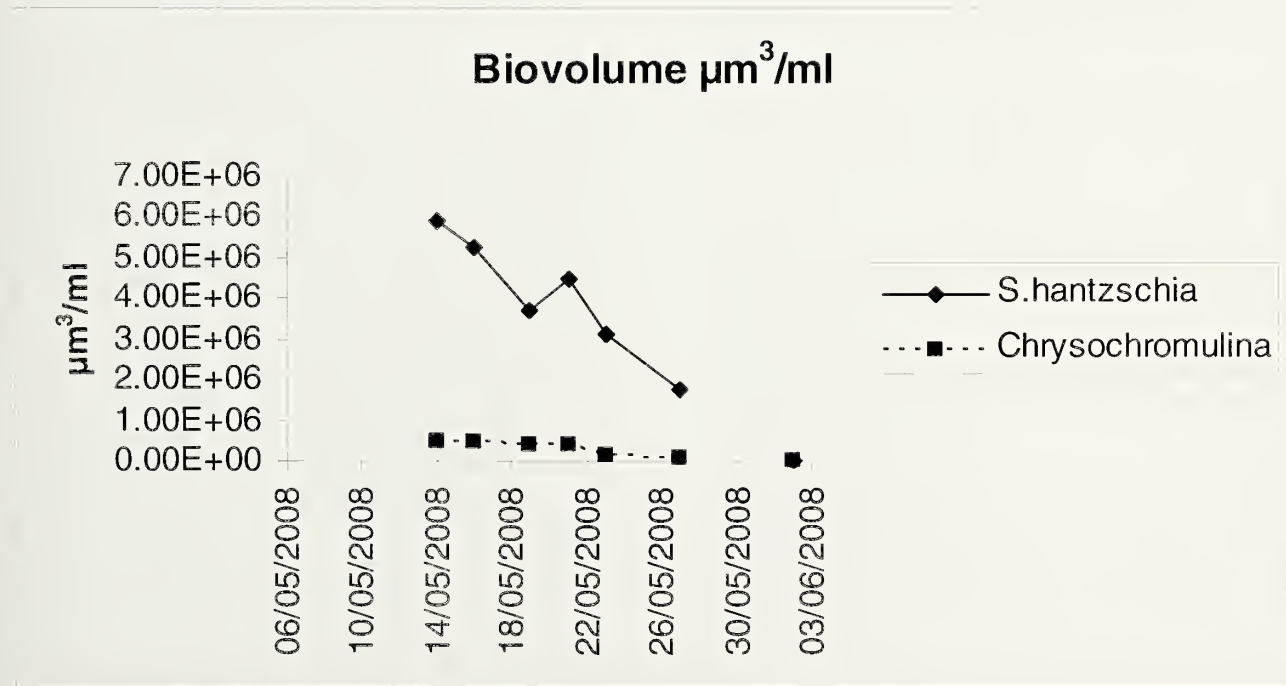
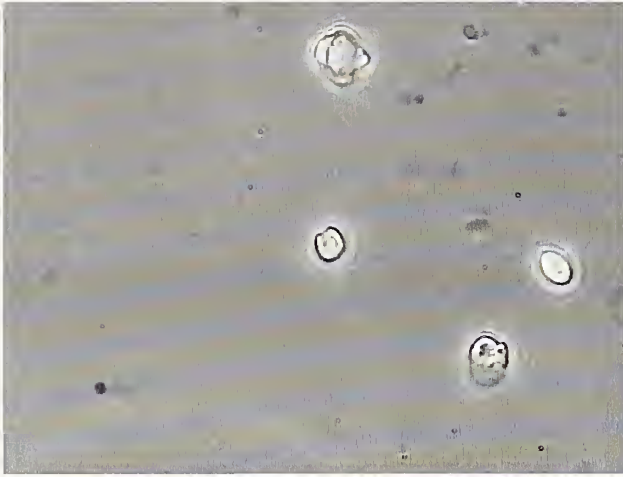
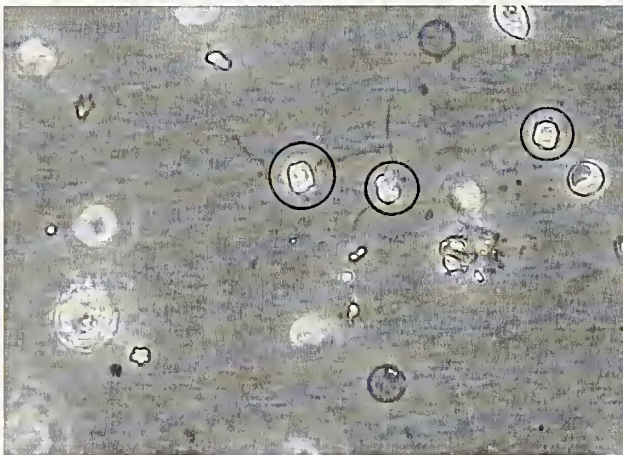


Fig. 3. Biovolume of the two dominant algae in Strathclyde Loch.



**Fig. 4a.** Live *Chrysochromulina parva* Lackey from Strathclyde Loch (14/05/2008), x400 mag., phase contrast, Leica DMIRB. The cell is 5  $\mu\text{m}$  in length, with a visible long straight haptonema 70  $\mu\text{m}$  long, and two flagella at either side 10-12  $\mu\text{m}$  long.



**Fig. 4b.** Same sample following preservation with Lugol's solution, showing distortion of *Chrysochromulina* cells (circled), flagella are visible but haptonema is hidden/coiled. x400 mag., phase contrast, Leica DMIRB.

## DISCUSSION

It is highly likely that *C. parva* was missed in the initial examination and was recorded as an unidentified flagellate nanophytoplankton due to the alga's extreme fragility, as the sample was not examined soon after collection. No further unidentified flagellate nanophytoplankton of the type were recorded in following examinations.

Toxic events associated with blooms of *Chrysochromulina* have been extensively reported primarily from marine waters and have led to extensive fish mortalities, and have also been associated with zooplankton, mollusc, sponge and crab mortalities. Unusually, toxic events associated with *Chrysochromulina* have also resulted in dead and dying red macroalgae *Delesseria sanguinea* (Smayda, 2006). Such reports of *Chrysochromulina*-associated fish mortalities are principally attributed to *C. polylepis*

and *C. leadbeateri*. Hansen *et al.* however, reported mortalities of caged rainbow trout in the southern Kattegat, Lillebaelt region of Denmark, associated with a mixed bloom of *C. brevifilum*, *C. ericina*, *C. hirta* and *C. spinifera*, with fish dying mainly during the early bloom stages when cell concentrations were between 10,000 and 30,000 cells/ml, with the maximum *Chrysochromulina* concentration of 50,000 cells/ml (Hansen *et al.*, 1995). Although the cause of the farmed fish mortalities was unclear, the effects on the fish suggest that one or several *Chrysochromulina* species may be toxic.

In freshwater systems, *C. parva* were mostly abundant in the summer but also common in spring, with densities of 20,000 cells/ml recorded in spring in a number of Lake District lakes and tarns (Parke *et al.*, 1962). Lund reported densities of 32,000 cells/ml, without associated fish mortalities in Esthwaite Water during June (Lund, 1961). Periodicity was not apparently driven by fluctuations in temperature or light. Kristiansen reported *C. parva* densities of 50,000 cells/ml also without associated fish mortalities (Kristiansen, 1971). Relatively higher densities of *C. parva* (614,000 cells/ml) were reported in a small Danish lake during June, with dead fish being reported from the beginning of May (Hansen *et al.*, 1994). This would have represented the first report of toxicity in a freshwater *Chrysochromulina* had toxicity tests been carried out.

In Scottish freshwater lochs, *Chrysochromulina* would appear to be a scarce component of the phytoplankton community occurring primarily during the spring and late summer, although the relative small size of the alga (diameter between 5-6  $\mu\text{m}$ ), its extreme fragility, and the difficulty associated with identifying it from preserved samples may have led to the alga being under-reported in fresh waters. Surveys of a number of Scottish freshwater lochs in 2007 indicated the presence of *Chrysochromulina* during July-September 2007 at low concentrations of between 10s-100s cells/ml, with concentrations of 1,019 cells/ml recorded from Loch Scadabhagh (North Uist) during September 2007, and 2,415 cells/ml from Loch Ussie (near Dingwall) in July 2007 (SEPA unpublished data). Analysis of a water sample from a small pond in Biggar (Lanarkshire) during October 2006 detected *Chrysochromulina* at densities of 4,215 cells/ml, with their presence again noted during May 2008. *Chrysochromulina* was also noted in Lochend Loch (North Lanarkshire) during May 2008 at densities of 1,447 cells/ml. There were no reported associated fish mortalities at any of these sites (SEPA unpublished information).

The abundance of *Chrysochromulina* in Strathclyde Loch, with maximum cell concentrations of 11,500 cells/ml during May, was at the lower range of concentrations reported so far from freshwaters, but within concentrations reported from marine waters where fish mortalities were detailed. The bloom during May 2007 could be regarded as a high



population density, low biomass bloom, consistent with previous blooms of *Chrysochromulina* (Smayda, 2006). High population density, low biomass blooms are regarded as harmful not because of their toxicity, but because their high population density and small size may lead to clogging of feeding appendages (Smayda, 1997), and perhaps also fish gills. The unexplained fish deaths at Strathclyde Loch could also be due to a combination of factors such as stress from the relatively high temperature, high dissolved oxygen and pH in the water, which all declined at the end of the sampling period. Irritation/clogging of fish gills with diatoms, and possibly bacterial exotoxins associated with die-off of the diatom bloom could be additional factors leading to fish mortalities (Environment Agency, 2003).

The abundance of *Chrysochromulina* however, although at relatively modest concentrations, and its past association with fish mortalities may suggest a toxic effect. The possibility of *Chrysochromulina* toxicity in Scottish freshwater lochs therefore deserves future monitoring.

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